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**Name of Organization:** Ohio State University Research Foundation

**Type of Organization:** College or University

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**Project Title:** Protection of the Genetic Integrity of Great Lake Esocids

**Project Category:** Habitat (Ecological) Protection and Rest

**Rank by Organization (if applicable):** 0

**Total Funding Requested (\$):** 103,479 **Project Duration:** 2 Years

**Abstract:**

This project aims to develop sterile tiger muskie strains for recreational sport fishery stocking programs in the Great Lakes region. Superior triploid tiger muskies should help protect native muskellunge or northern pike fisheries from accidental or stocking because the presumed sterility of diploid tiger muskies has been shown to be false (Black and Williamson 1946. Wisc. Acad. Sci. Arts and Lett. 38: 299-314; Casselman et al 1986; Am. Fish. Soc. Spec. Publ. 15: 14-46).

We will produce 100% triploid tiger muskies using a pressure shock of 8000 psi applied at a time of initiation of 15 min for a duration of 10 min. These parameters have been previously optimized in our laboratory. We aim to develop molecular markers that can be used to distinguish the tiger muskie hybrids from the original parental species. We will develop species-specific and parent-specific markers for our fish populations by screening two types of PCR-based genetic markers: RAPDs and microsatellites. RAPD markers will be screened using standard techniques (e.g., Van Eenennaam et al. 1996. Aquaculture 147:177-189). We will also screen the species-specific microsatellite marker Elu25 (Miller and Kapuscinski 1996. Trans. Am. Fish. Soc 125: 971-977). If needed, we will develop additional microsatellite markers and screen allozyme markers. We will develop at least three reliable genetic markers that can be used to verify hybrids. We will distinguish the diploid from triploid hybrids using flow cytometry. Hybrids will be identified using at least three molecular genetic markers (described above). We will also attempt to use the rarely observed "silver pike" trait, a color morph of northern pike recently found in some Ontario lakes (Foster and Somerville 2000. Fifty-sixth Annu. Northeast Fish and Wildl. Conf., Charleston, VA). The ploidy of the progeny will be determined using flow cytometry (Lin and Dabrowski 1996. Can. J. Fish. Aquat. Sci. 53: 2067-2075).

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**Geographic Areas Affected by the Project**

**States:**

<input type="checkbox"/> Illinois	<input type="checkbox"/> New York
<input type="checkbox"/> Indiana	<input type="checkbox"/> Pennsylvania
<input type="checkbox"/> Michigan	<input type="checkbox"/> Wisconsin
<input type="checkbox"/> Minnesota	<input checked="" type="checkbox"/> Ohio

**Lakes:**

<input type="checkbox"/> Superior	<input checked="" type="checkbox"/> Erie
<input type="checkbox"/> Huron	<input type="checkbox"/> Ontario
<input checked="" type="checkbox"/> Michigan	<input type="checkbox"/> All Lakes

**Geographic Initiatives:**

<input type="checkbox"/> Greater Chicago	<input checked="" type="checkbox"/> NE Ohio	<input checked="" type="checkbox"/> NW Indiana	<input checked="" type="checkbox"/> SE Michigan	<input checked="" type="checkbox"/> Lake St. Clair
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**Primary Affected Area of Concern:**

**Other Affected Areas of Concern:**

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***For Habitat Projects Only:***

**Primary Affected Biodiversity Investment Area:**

**Other Affected Biodiversity Investment Areas:**

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**Problem Statement:**

Although only purebred muskies are stocked in Ohio waters, the threat of accidental or intentional stocking of hybrid tiger muskies is real. Neighboring states such as Michigan, Indiana, and Pennsylvania use tiger muskies in their stocking programs. Thus, tiger muskies that are stocked in these states may stray into Lake Erie waters, potentially leading to genetic contamination of the native muskellunge populations. Molecular markers will allow us to quickly and accurately distinguish tiger muskies from their muskellunge or northern pike parents.

**Proposed Work Outcome:**

Diploid and triploid tiger muskies are currently maintained at the aquaculture laboratory of Dr. Dabrowski and these fish will be used for observation of sex ratio, gametogenesis and gamete fertility. These fish can be used for the development of species-specific molecular markers but not for the development of parent-specific markers because tissue samples from the parents of these hybrids were not obtained. Thus, additional triploid tiger muskies will be produced in two subsequent years of this project using optimized conditions of pressure shock, time after activation, and duration. Fin and tissue samples from diploid and triploid tiger muskie groups, including their parents will be obtained. Muskellunge will be sampled in Clear Fork Reservoir, Ohio. Screening of molecular markers for distinguishing the F1 hybrids from their parental species will be carried out in conjunction with an attempt to use the "silver pike" trait from Ontario lakes as color marker.

We aim to develop molecular markers that can be used to distinguish the hybrids from the original parental species. We will develop species-specific markers for our fish populations by screening two types of PCR-based genetic markers: RAPDs and microsatellites. RAPD markers will be screened using standard techniques (e.g., Van Eenennaam et al. 1996; Aquaculture 147: 177-189). We will also screen the species-specific microsatellite marker Elu25 (Miller and Kapuscinski 1996. Trans.Am. Fish. Soc. 125: 971-977). If needed, we will develop additional microsatellite markers and screen allozyme markers. We will develop at least three reliable genetic markers that can be used to verify hybrids.

We will distinguish the diploid from triploid hybrids using flow cytometry (Lin and Dabrowski 1996. Can. J. Fish. Aquat. Sci. 53: 2067-2075 ; Garcia-Abiado et al. 1999. North Am. J. Aquaculture 61:319-325 ). Hybrids will be identified using at least three molecular genetic markers (described above).

In the first year of the project, we will concentrate on the hybrid between female muskie and male northern pike. In the second year, the reciprocal cross will be examined.

In many states, annual stocking programs focus on tiger muskies because they are easier to raise on dry diets, easier to

catch than muskellunge, and because they grow faster than northern pike. Tiger muskies are more tolerant to higher water temperatures (>28C) and much more resistant to infections and diseases (our own observations). Despite the widespread use of diploid tiger muskies, triploids should be the only alternative considered for stocking because they would be unable to interbreed with native fish. Reports of sterility in tiger muskies may be inaccurate because very few fish (females) were examined for fertility. Thus, the possibility that introductions of fertile tiger muskies might lead to genetic contamination of native fish populations could be avoided by stocking triploid, rather than diploid tiger muskies. Triploid tiger muskies should have better survival and growth, and should be completely sterile compared to the diploids. Thus, the use of triploid tiger muskies in annual stocking programs should result in lower adverse genetic and ecological impacts on native fish populations and Great Lakes ecosystems.

**Project Milestones:**

**Dates:**

Project Start	05/2000
Production of triploid tiger muskie	05/2000
Collection of tissue samples	05/2000
Sampling of purebred muskellunge	05/2000
Begin molecular screening experiments	06/2000
Production of triploid tiger muskie	05/2001
Collection of tissue samples	05/2001
Project End	05/2002

☐ Project Addresses Environmental Justice

**If So, Description of How:**

☒ Project Addresses Education/Outreach

**If So, Description of How:**

The results obtained from this study will be published in peer-reviewed journals dealing with aquaculture and/or fisheries management. News articles will also be published in local and regional newspapers and in the Ohio Sea Grant newsletter "Twine Line". Findings will also be made available in the web site of the School of Natural Resources, The Ohio State University and the Ohio State Information Center which prepares fact sheets and press releases.

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**Project Budget:**

	<b>Federal Share Requested (\$)</b>	<b>Applicant's Share (\$)</b>
<b>Personnel:</b>	31,642	25,000
<b>Fringe:</b>	7,752	0
<b>Travel:</b>	4,000	0
<b>Equipment:</b>	0	0
<b>Supplies:</b>	25,000	0
<b>Contracts:</b>	0	0
<b>Construction:</b>	0	0
<b>Other:</b>	2,000	0
<b>Total Direct Costs:</b>	70,394	25,000
<b>Indirect Costs:</b>	33,085	0
<b>Total:</b>	103,479	25,000
<b>Projected Income:</b>	0	0

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**Funding by Other Organizations (Names, Amounts, Description of Commitments):**

None

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**Description of Collaboration/Community Based Support:**

We will collaborate with the Ohio Department of Natural Resources, Division of Wildlife, in collecting tissue samples from native muskellunge and northern pike populations. Dr. Konrad Dabrowski will collaborate with Dr. Glenn Howe, Assistant professor of Forest Genetics at the School of Natural Resources, Wooster Campus and with Dr. Mary Ann Garcia-Abiado, postdoctoral researcher at the School of Natural Resources, The Ohio State University Columbus campus. Dr. Howe is an expert in molecular genetics and Dr. Garcia-Abiado is an expert on ploidy manipulation and sex control in fish.